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GAS CHROMATOGRAPHY

Gas chromatography (GC) can also be referred to as **vapor-phase chromatography** (VPC) and even **gas-liquid chromatography** (GLC). Usually, the technique, the instrument, and the chart recording of the data are called **GC**:

<i>"Fire up the GC."</i>	(the instrument)
<i>"Analyze your sample by GC."</i>	(perform the technique)
<i>"Get the data off the GC."</i>	(analyze the chromatogram)

I've mentioned the similarity of all chromatography, and just because electronic instrumentation is used, there's no need to feel that something basically different is going on.

THE MOBILE PHASE: GAS

In **column chromatography** the mobile (moving) phase is a liquid that carries your material through an adsorbent. I called this phase the **eluent**, remember? Here a gas is used to push, or *carry*, your vaporized sample, and it's called the **mobile phase**.

The **carrier gas** is usually helium, although you can use nitrogen. You use a **microliter syringe** to inject your sample into this gas stream through an **injection port**, then *onto the column*. If your sample is a mixture, the *compounds separate on the column* and reach the **detector** at different times. As each component hits the detector, the detector generates an **electric signal**. Usually, the signal goes through an **attenuator network**, and then out to a **chart recorder** to record the signal. I know, it's a fairly general description and Fig. 137 is a highly simplified diagram, but there are lots of different GCs, so being specific about their operation doesn't help here. You should see your instructor. But that doesn't mean we can't talk about some things.

GC SAMPLE PREPARATION

Sample preparation for GC doesn't require much more work than handing in a sample to be graded. *Clean and dry*, right? Try to take care that the boiling point of the material is low enough that you can actually work with the technique. The maximum temperature depends on the type of column,

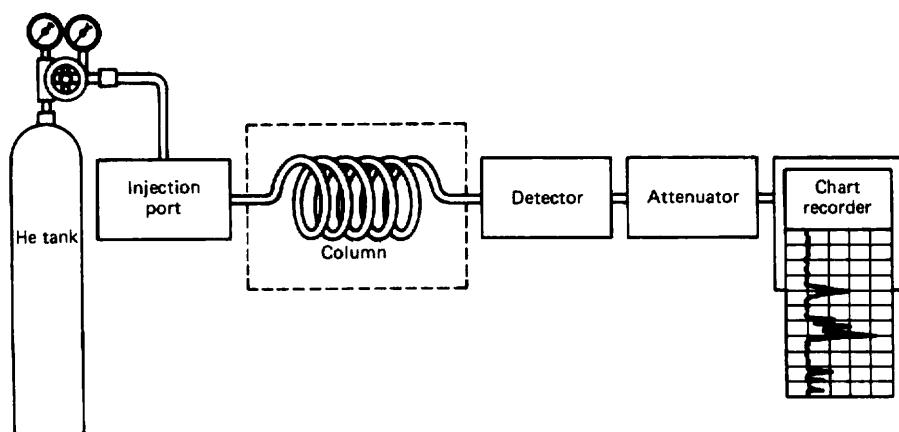


Fig. 137 Schematic of a gas-chromatography setup.

and that should be given. In fact, for any single experiment that uses GC, the nature of the column, the temperature, and most of the electronic settings will be fixed.

GC SAMPLE INTRODUCTION

The sample enters the GC at the **injection port** (Fig. 138). You use a microliter syringe to pierce the rubber septum and inject the sample onto the column. Don't stab yourself or anyone else with the needle. Remember,

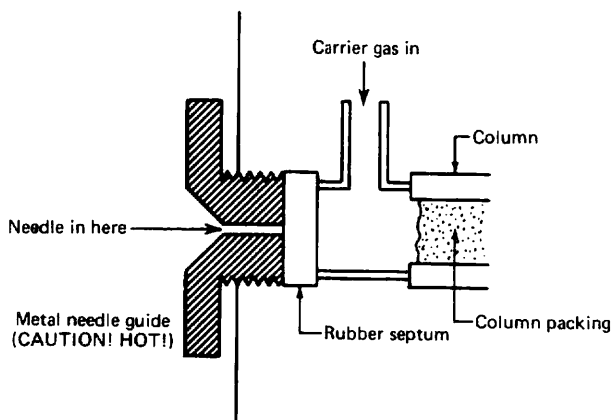


Fig. 138 A GC injection port.

this is *not* dart night at the pub. Don't throw the syringe at the septum. There is a way to do this.

1. To load the sample, put the needle into your liquid sample and *slowly* pull the plunger to draw it up. If you move too fast, and more air than sample gets in, you'll have to push the plunger back again and draw it up once more. Usually, they give you a 10[m>l syringe, and 1 or 2[m>l of sample is enough. Take the loaded syringe out of the sample, and *carefully, cautiously*, pull the plunger back so there is no sample in the needle. You should see a bit of air at the very top but not very much. This way you don't run the risk of having your compound boil out of the needle as it enters the injector oven just before you actually inject your sample. That broadens the sample and reduces the resolution. In addition, the air acts as an internal standard. Since air travels through the column almost as fast as the carrier gas, the **air peak** that you get can signal the start of the chromatogram, much like the notch at the start of a TLC plate. Ask your instructor.
2. Hold the syringe in *two hands* (Fig. 139). There is no reason to practice being an M.D. in the organic laboratory.
3. Bring the syringe to the level of the injection port, straight on. No angles. Then let the needle touch the septum at the center.
4. The real tricky part is holding the barrel and, without injecting, pushing the needle through the septum. This is easier to write about than it is to do the first time.
5. Now quickly and smoothly push on the plunger to inject the sample, and pull the syringe needle out of the septum and injection port.

After a while, the septum gets full of holes and begins to leak. Usually, you can tell you have a leaky septum when the pen on the chart recorder wanders about aimlessly without any sample injected.

SAMPLE IN THE COLUMN

Now that the sample is in the column, you might want to know what happens to this mixture. Did I say mixture? Sure. Just as with **thin-layer** and **column chromatography**, you can use GC to determine the composition or purity of your sample.

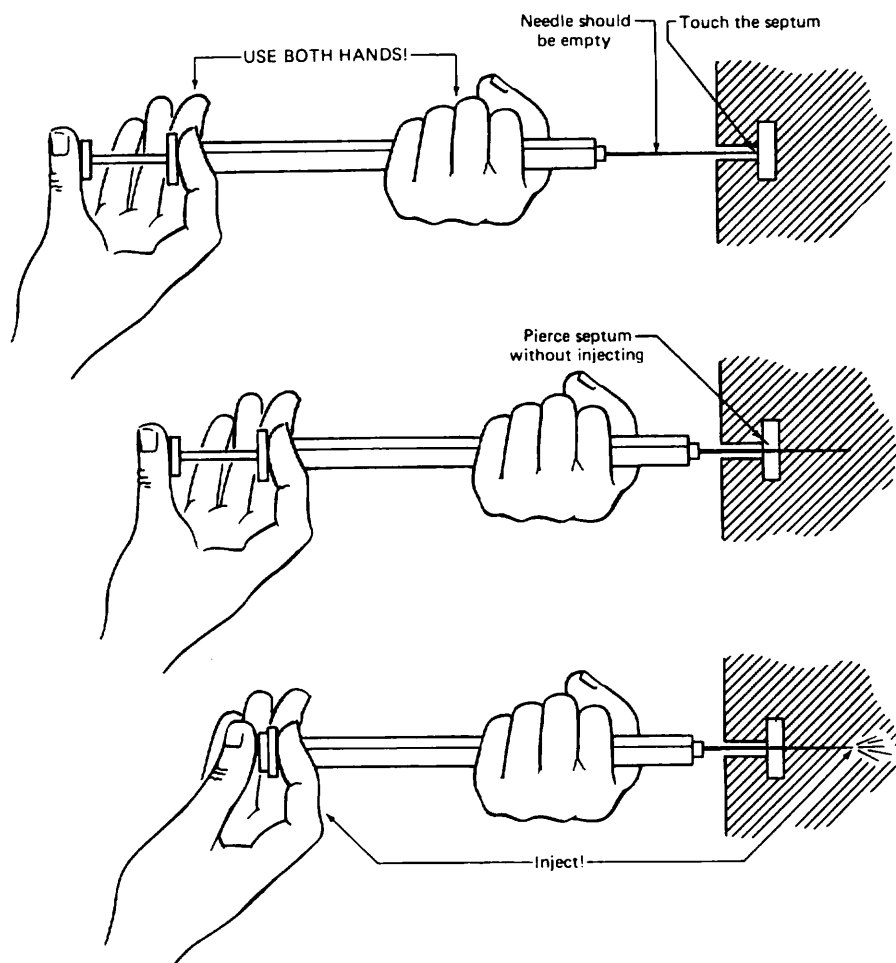


Fig. 139 Three little steps to a great GC.

Let's start with two components, A and B again, and follow their path through an **adsorption column**. Well, if A and B are different, they are going to stick on the adsorbent to different degrees and spend more or less time flying in the carrier gas. Eventually, one will get ahead of the other. *Aha! Separation*—just like column and thin-layer chromatography. Only here the samples are vaporized, and it's called **vapor-phase chromatography (VPC)**.

Some of the adsorbents are coated with a **liquid phase**. Most are very high-boiling liquids, and some look like waxes or solids at room tempera-

ture. Still, they're liquid phases. So, the different components of the mixture you've injected will spend different amounts of time in the liquid phase and, again, you'll get a separation of components in your compound. Thus the technique is known as **gas-liquid chromatography (GLC)**. Thus you could use the same adsorbent and different liquid phases, and change the characteristics of each column. Can you see how the sample components would partition themselves between the gas and liquid phases and separate according to, perhaps, molecular weight, polarity, size, and so on, making this technique also known as **liquid-partition chromatography**?

Since these liquid phases on the adsorbent are eventually liquids, you can boil them. And that's why there are temperature limits for columns. It is not advisable to heat a column past the recommended temperature, boiling the liquid phase right off the adsorbent and right out of the instrument.

High temperature and air (oxygen) are death for some liquid phases, since they oxidize. So make sure the carrier gas is running through them at all times, even a tiny amount, while the column is hot.

SAMPLE AT THE DETECTOR

There are several types of **detectors**, devices that can tell when a sample is passing by them. They detect the presence of a sample and convert it to an electrical signal that's turned into a **GC peak** (Fig. 140) on the chart recorder. The most common type is the **thermal conductivity detector**. Sometimes called hot-wire detectors, these devices are very similar to the filaments you find in light bulbs, and they require some care. Don't ever turn on the **filament current** unless the carrier gas is flowing. A little air (oxygen), a little heat, a little current, and you get a lot of trouble replacing the burned-out detector.

Usually, there are at least two thermal conductivity detectors in the instrument, in a "bridge circuit." Both detectors are set in the gas stream, but only one gets to see the samples. The electric current running through them heats them up, and they lose heat to the carrier gas at the same rate.

As long as no sample, only carrier gas, goes over *both* detectors, the bridge circuit is **balanced**. There is no signal to the recorder, and the pen does not move.

Now a sample in the carrier gas goes by one detector. This sample has a thermal conductivity different from that of pure carrier gas. So the **sample detector** loses heat at a different rate from the **reference detector**.

Height (mm)	8	68	145
Width at half-height (mm)	4	4	4
Area (mm ²)	32	212	580
Relative area	1	8.5	18.1
Distance from injection (in.)	3	3 $\frac{5}{16}$	3 $\frac{1}{2}$
Retention times t_R at 2 min/in. (min)	6	6.63	7.38

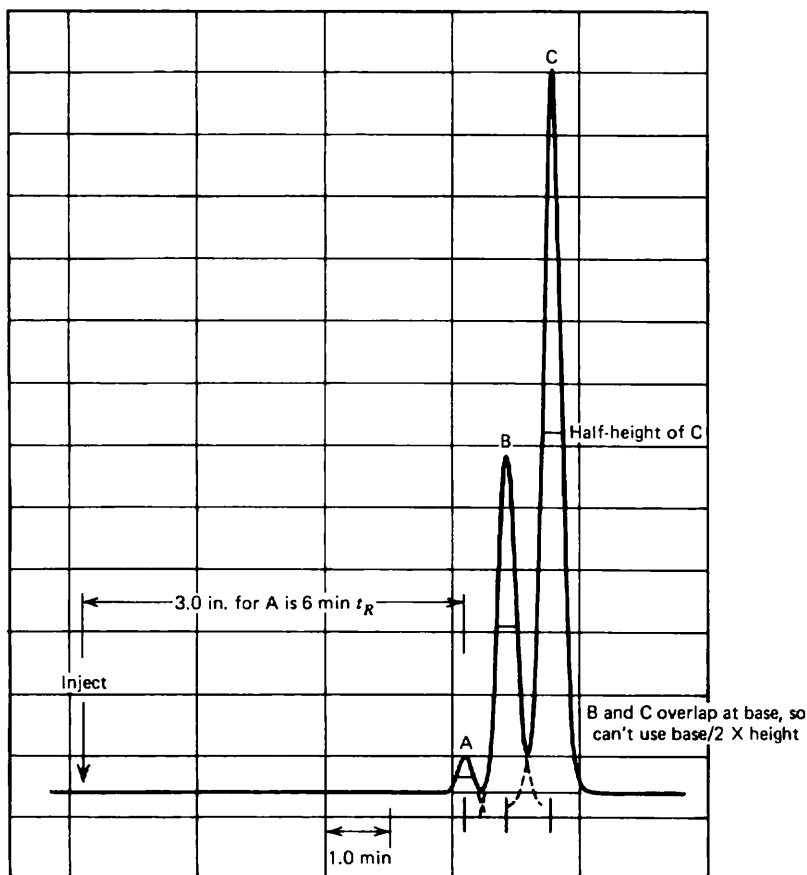


Fig. 140 A well-behaved GC trace showing a mixture of three compounds.

(Remember, the **reference** is the detector that NEVER sees samples—only carrier gas.) The detectors are in different surroundings. They are not really equal any more. So the bridge circuit becomes unbalanced, and a signal goes to the chart recorder, giving a GC peak.

Try to remember the pairing of **sample** with **reference**; it's the *difference* in the two that most electronic instrumentation responds to. You will see this again and again.

ELECTRONIC INTERLUDE

The electrical signal makes two other stops on its way to the chart recorder.

1. **The coarse attenuator.** This control makes the signal weaker (attenuates it). Usually, there's a scale marked in **powers of two**: 2, 4, 8, 16, 32, 64, So each position is half as sensitive as the last one. There is one setting, either an [8] or an **S** (for **shorted**), which means that the attenuator has *shorted out the terminals connected to the chart recorder*. Now the **chart recorder zero** can be set properly.
2. **The GC zero control.** This control helps set the zero position on the chart recorder, but it is *not to be confused with the zero control on the chart recorder*.

Here's how to set up the electronics properly for a GC and a chart recorder.

1. The **chart recorder** and **GC** should be allowed to warm up and stabilize for at least 10–15 minutes. Some systems take more time; ask your instructor.
2. Set the **coarse attenuator** to the highest attenuation, usually an [8] or an S.
3. Now set the pen on the chart recorder to zero using *only the chart recorder zero control*. Once you do that, *leave the chart recorder alone*.
4. Start turning the *coarse attenuator* control to more sensitive settings (lower numbers) and *watch the pen on the chart recorder*.
5. If the pen on the chart recorder moves off zero, *use the GC zero control only* to bring the pen back to the zero line on the chart recorder paper.
6. *Do not touch the chart recorder zero. Use the GC zero control only.*
7. As the *coarse attenuator* gets to more sensitive settings (lower numbers), it becomes more difficult to adjust the *chart recorder pen to zero* using *only the GC zero control*. Do the best you can at the *lowest attenuation* (highest sensitivity) you can hold a zero steady at.
8. Now, you *don't* normally run samples on the GC at attenuations of 1 or 2. These settings are *very sensitive*, and there may be lots of electrical noise—the pen jumps about. The point is, *if the GC zero is OK at an attenuation of 1*, then when you run at attenuations of 8, 16, 32, and so on, *the baseline will not jump if you change attenuation in the middle of the run*.

Now that the attenuator is set to give peaks of the proper height, you're ready to go. Just be aware that there may be a **polarity switch** that can make your peaks shift direction.

SAMPLE ON THE CHART RECORDER

Interpreting a GC is about the same as interpreting a TLC plate, so I'll use TLC terms as a comparison to show the similarities. Remember the R_f value from TLC? The ratio of the distance the eluent moved to how far the spots of compound moved? Well, distances can be related to times, so the equivalent of R_f in GC is **retention time**. It's the time it takes the sample to move through the column minus the time it takes for the carrier gas to move through the column. Remember the part about putting air into the syringe to get an **air peak**? Well, you can assume that air travels with the carrier gas and doesn't interact with the column material. So the air peak that shows up on the chart paper can be considered to be the reference point, the "notch," as it were, that marks the start, just as on the TLC plates.

OK, so you don't want to use an air peak. Then make a mark on the chart paper as soon as you've injected the sample, and use *that* as the start. It's not as good, but it'll work.

No. You do not need a stopwatch for the retention times. Find out the distance the chart paper crawls in, say, a minute. Then get out your little ruler and measure the distances from the starting point (either air peak or pen mark) to the midpoint of each peak on the baseline (Fig. 140). Don't be wise and do any funny angles. It won't help. You've got the distances and the chart speed, so you've got the retention time. It works out. Trust me.

You can also estimate how much of each compound is in your sample by measuring **peak areas**. The area under each GC peak is proportional to the amount of material that's come by the detector in that fraction. You might have to make a few assumptions (e.g., the peaks are truly triangular, and each component gives the same response at the detector), but usually it's pretty straightforward. Multiply the height of the peak by the width at half the height. If this sounds suspiciously like the area of a triangle, you're on the right track. It's usually not half the base times the height, however, since sometimes the **baseline** is not very even, and that measurement is difficult.

PARAMETERS, PARAMETERS

To get the best GC trace from a given column, there are lots of things you can do, simply because you have so many controls. Usually, you'll be told the correct conditions, or they'll be preset on the GC.

Gas Flow Rate

The faster the carrier gas flows, the faster the compounds are pushed through the column. Because they spend *less time in the stationary phase*, they don't separate as well, and the GC *peaks come out very sharp but not well separated*. If you *slow the carrier gas down too much*, the compounds spend so much time in the stationary phase that *the peaks broaden and overlap gets very bad*. The optimum is, as always, the best separation you need, in the shortest amount of time. Sometimes you're on your own. Most of the time, someone else has already worked it out for you.

Temperature

Whether or not you realize it, the GC column has its own heater—the **column oven**. If you turn the temperature up, the compounds hotfoot it through the column very quickly. Because they spend *less time in the stationary phase*, they don't separate as well, and the GC *peaks come out very sharp but not well separated*. If you *turn the temperature down some*, the compounds spend so much time in the stationary phase that *the peaks broaden and overlap gets very bad*. The optimum is, as always, the best separation you need in the shortest amount of time. There are two absolute limits, however.

1. *Too high a temperature, and you destroy the column.* The adsorbent may decompose, or the liquid phase may boil out onto the detector. *Never exceed the recommended maximum temperature for the column material.* Don't even come within 20°C of it.
2. *Way too low a temperature, and the material condenses on the column.* You have to be above the **dew point** of the least volatile material. Not the boiling point. Water doesn't always condense on the grass—

become dew—every day that's just below 100°C (that's 212°F, the boiling point of water). Fortunately, you don't have to know the dew points for your compounds. However, you *do* have to know that you *don't have to be above the boiling point of your compounds*

Incidentally, the **injector** may have a separate **injector oven**, and the **detector** may have a separate **detector oven**. Set them both 10 to 20°C higher than the column temperature. You can even set these *above the boiling points* of your compounds, since you *do not want them to condense in the injection port or the detector, ever*. For those with *only one temperature control*, sorry. The injection port, column, and detector are all in the same place, all in the same oven, and all at the same temperature. The *maximum temperature*, then, is *limited by the decomposition temperature of the column*. Fortunately, because of that dew point phenomenon, you really don't have to work at the boiling points of the compounds either.